

Amendments to the Specification:

Please add the following paragraph between lines 3 and 4 on page 1.

C1 This application claims the benefit of United States provisional application 60/162,491 filed October 29, 1999, which is hereby incorporated by reference herein in its entirety.

Please replace the paragraph on page 9, lines 13-20 with the following paragraph:

C2 **Figure 8** Figures 8A-8D [[shows]] show octa-MAP1-induced IgG anti-LOS antibody responses in mice. (A) Eight mice received a dose of 50 µg of Octa-MAP1 emulsified in Freund's adjuvant on day 0 and again on day 21. (B) Four mice were immunized with purified LOS as a positive control. Mice were immunized with either Freund's adjuvant (C) or an unrelated octa-MAP control peptide (D) as negative controls.

Please replace the paragraph on page 16, line 20 to page 17 line 9 with the following paragraph:

C3 Positive *E. coli* clones were grown overnight in IMC media containing 100 µg/ml ampicillin, at 25°C and then induced to express the peptide fusions for 6 h. *E. coli* cells were fixed with 0.5% paraformaldehyde on ice for 10 min. Aliquots of 200-µl of fixed organisms were spun at 2000 X g for 10 min. Supernatants were discarded, and pellets were resuspended in blocking buffer (IMC media containing 100 µg/ml ampicillin, 1% nonfat dry milk, 150 mM NaCl and 1% α - methyl mannoside) containing mAb 2C7. Suspensions were incubated at 37°C for 30 min. before spinning at 2000 X g for 10 min. Pellets were washed with 100 µl of washing buffer (IMC media containing 100µg/ml ampicillin and 1% α-methyl mannoside) and then resuspended in 100 µl of blocking buffer containing FITC-conjugated anti-mouse IgG (Sigma, St. Louis, MO). The mixtures were incubated at 37°C for 30 min before spinning at 2000 X g for 10 min. Supernatants were removed, and pellets washed in 100 µl of washing buffer before resuspension in 1 ml of PBS. The suspensions were analyzed on a FACS using CellQuest software CELLQUEST™ Flow Cytometry Software (Becton Dickinson, Franklin Lakes NJ). A negative clone that did not bind mAb 2C7 was used as a control.

Please replace the paragraph on page 18, lines 13-22 with the following paragraph:

C4 A synthetic peptide (PEP1; IPVLDENGLFAP [SEQ ID NO:1]) whose sequence corresponds to the consensus sequence "DE_GLF" and includes two cysteine flanking regions (CGP- and -GPC residues at the [[N]]- and C- terminus, respectively) was synthesized (Boston Biomolecules, MA) to assess specific binding to 2C7 mAb by inhibition ELISA and to determine whether peptide mimics characterized as thioredoxin-fusion proteins would retain the antigenicity independent of the fusion context [SEQ ID NO:10].

Please replace the paragraph on page 18, line 31 to page 19 line 14 with the following paragraph:

C5
Peptides were diluted in blocking buffer (1% ovalbumin, 0.05% ~~tween-20~~ TWEEN-20TM (polysorbate 20), 0.5 M NaCl in PBS) to produce mixtures of varying concentrations (0.1, 0.5 and 1 mg/ml). 50 µl-aliquots from each of the concentrations were incubated with 50 µl of mAb 2C7 (stock concentration 2 µg/ml diluted in blocking buffer) at 37°C for 1 h, then 100 µl of the mixtures were loaded into microtiter plate wells coated with purified LOS prepared from strain 15253 (80 µg/ml). The wells were incubated at 37°C for 1 h, then washed. After the wells were washed, bound mAb 2C7 was detected with anti-mouse IgG conjugated to alkaline phosphatase. Purified LOS prepared from gonococcal strain 15253 was used as a positive control. A non-reactive 15-mer peptide sequence generated by the above described random peptide library system was used as a negative control peptide [SEQ ID NO:9].

Please replace the paragraph on page 20, line 23 to page 21, line 10 with the following paragraph:

C6
Solid phase ELISA was performed to assess the binding of mAb 2C7 to multiple antigen peptides. For direct ELISA, Immulon 1 plates were coated overnight with multiple antigen peptides (1 µg/well) and reacted with varying concentration of mAb 2C7. For inhibition ELISA, plates were coated with purified LOS prepared from *N. gonorrhoeae* strain 15253 (80 µg/ml) at 37°C for 3 h. Peptides (linear or MAPs) were diluted in blocking buffer (1% ovalbumin, 0.05% ~~tween-20~~ TWEEN-20TM (polysorbate 20), 0.5 M NaCl in PBS) to produce mixtures of varying concentrations. 50 µl-aliquots from each concentration were incubated with 50 µl of mAb 2C7 (stock concentration 0.4 µg/ml diluted in blocking buffer) at 37°C for 1 h, then 100 µl of mixtures were loaded into microtiter plate wells. The wells were incubated at 37°C for 1 h, then washed. After the wells were washed, bound mAb 2C7 was detected with anti-mouse IgG conjugated to alkaline phosphatase. Purified LOS prepared from gonococcal strain 15253 was used as a positive control in inhibition ELISA.

Please replace the paragraph on page 21, line 23 with the following paragraph:

C7
Immunization with octa-MAP1 induces an IgG anti-LOS antibody response in mice, as shown in ~~Figure 8~~ Figures 8A-8D. The response profile seen in Figure 8(A), in which there is no significant IgG anti-LOS response until the boost at week 3, indicates that the Octa-MAP1 elicited a T-cell dependent immune response in the responding mice. These results demonstrate the promise of a peptide mimic, such as Octa-MAP1, for immunizing humans against *N. gonorrhoeae* infection.

Please replace the paragraph on page 21, line 32 with the following paragraph:

C8
In Figure 8(A), eight mice received a dose of 50 µg of Octa-MAP1 emulsified in Freund's adjuvant on day 0 and again on day 21. Octa-MAP1, which mimics the 2C7 oligosaccharide epitope, induced IgG anti-LOS antibody in three of the eight mice. IgG

C8 control
anti-LOS responses in these three mice rose significantly after the first boost at week 3, peaked at week 7 (the next time measured) and decreased thereafter. Figure 8(B) shows the positive control experiment in which four mice were immunized with purified LOS. In these mice, IgG anti-LOS titers increased minimally after the first immunization and rose after boosting. All mice in the LOS group showed an anti-LOS antibody response. Four mice immunized with either Freund's adjuvant (C) or an unrelated octa-MAP control peptide (D), both negative controls, elicited weak or no IgG anti-LOS responses. The mean IgG anti-LOS antibody responses from all immunized mice (from the experiments depicted in ~~Figure 8~~ Figures 8A-8D) are shown in Figure 9 (mean \pm SE, including animals that exhibited no response).

Please replace the paragraph on page 22, line 18 with the following paragraph:

C^h
IgG anti-LOS antibody responses for the responder mice only (from the experiments depicted in ~~Figure 8~~ Figures 8A-8D) are shown in Figure 10. Antibody response is defined as IgG anti-LOS (mean \pm SE) greater than 0.4 mg/ml (4 fold above baseline IgG anti-LOS levels). At 7 and 10 weeks after primary immunization, responder mice immunized with Octa-MAP1 elicited IgG anti-LOS antibody levels higher ($p < 0.001$) than antibody levels elicited by negative control antigens (Freund's adjuvant alone or unrelated octa-MAP control peptide).

Please replace the paragraph on page 22, line 28 with the following paragraph:

C¹⁰
IgM anti-LOS antibody responses for responder mice only (from the experiments depicted in ~~Figure 8~~ Figures 8A-8D) are shown in Figure 11. Mice immunized with Octa-MAP1 that had elicited IgG anti-LOS responses failed to respond with IgM anti-LOS levels higher than mice immunized with negative control antigens. Immunization with LOS (positive control) elicited IgM anti-LOS antibody levels higher than animals immunized with either Octa-MAP1 or negative control antigens (Freund's adjuvant alone or unrelated octa-MAP control peptide).